

Original Article



Cow's milk oral immunotherapy in real life: 8-year long-term follow-up study

Inês Mota, Susana Piedade, Ângela Gaspar , Filipe Benito-Garcia, Graça Sampaio, Luís Miguel Borrego, and Mário Morais-Almeida

Immunoallergy Department, CUF Descobertas Hospital, Lisbon, Portugal

OPEN ACCESS

Received: Nov 1, 2017
Accepted: Jul 13, 2018

*Correspondence to

Ângela Gaspar

Immunoallergy Department, CUF Descobertas Hospital, R. Mário Botas, 1998-018 Lisbon, Portugal.

Tel: +351-210025200

Fax: +351-210025220

E-mail: angela.gaspar@sapo.pt

Copyright © 2018. Asia Pacific Association of Allergy, Asthma and Clinical Immunology.

This is an Open Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (<https://creativecommons.org/licenses/by-nc/4.0/>) which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

ORCID iDs

Ângela Gaspar 

<https://orcid.org/0000-0001-8330-8016>

Author Contributions

Conceptualization: Susana Piedade, Ângela Gaspar, Mário Morais-Almeida. Data curation: Inês Mota, Susana Piedade, Ângela Gaspar, Filipe Benito-Garcia, Graça Sampaio, Luís Miguel Borrego. Formal analysis: Inês Mota, Luís Miguel Borrego. Investigation: Inês Mota. Supervision: Susana Piedade, Ângela Gaspar, Mário Morais-Almeida. Validation: Inês Mota. Writing - original draft: Inês Mota, Ângela Gaspar. Writing - review & editing: Susana Piedade, Ângela Gaspar, Luís Miguel Borrego, Mário Morais-Almeida.

ABSTRACT

Background: Oral immunotherapy (OIT) has been recognized as a promising treatment for severe and long-lasting cow's milk (CM) allergy. Once maintenance has been achieved, patients should maintain daily intake of CM to ensure desensitization. Clinical experience concerning long-term follow-up is scarce.

Objective: The authors aimed to assess long-term efficacy and safety of a maintenance phase of OIT in real life.

Methods: Prospective study of all children and adolescents, who underwent CM-OIT and were subsequently followed at our allergy center on maintenance dose (200 mL daily) for at least 36 months after reaching the maintenance phase (from 2009 to 2016).

Results: Forty-two patients were enrolled: 60% male, 36% with history of anaphylaxis and 57% with asthma. The median time of follow-up was 69 months (range, 39–105 months) and the median age at the last clinical evaluation was 13 years (range, 6–23 years). Regarding adherence to the protocol: 92% are on free diet (at least 200 mL of CM daily; 7-g protein); 14% had transient interruptions and 7% definitely withdrawn with loss of tolerance. During maintenance, 45% developed mild to severe allergic reactions, and 7% had more than 3 episodes. A positive correlation between the occurrence of allergic reactions and history of anaphylaxis ($p < 0.001$) was found. The coexistence of asthma was risk factor for the occurrence of allergic reactions during maintenance.

Conclusion: This real-life study supports long-term efficacy and safety of CM-OIT. Despite daily intake, 41% had symptoms at some moment during the complete follow-up period; a total of 33 symptomatic days in patients with mean follow-up time of 67.5 months. Clinical tolerance depends on daily intake. The protective effect reached can be lost after CM withdrawal. History of anaphylaxis was a risk factor for the occurrence of allergic reactions during the maintenance phase.

Keywords: Anaphylaxis; Cow's milk allergy; Food allergy; Oral desensitization; Oral immunotherapy; Specific oral tolerance induction

INTRODUCTION

Cow's milk (CM) is one of the major causes of food allergy in childhood. In a systematic review and meta-analysis of European studies, the self-reported lifetime prevalence of CM allergy (CMA) was 6% [1]. Retrospective studies showed that the prognosis is worse than previously reported, with increasing rates of CMA persistence up to adolescence [2]. Strict avoidance might be difficult since the widespread use of CM in processed food, particularly in those patients who react even with very small amounts of CM.

Oral immunotherapy (OIT), an innovative strategy, has been recognized as promising, effective and reasonably safe treatment for severe and long-lasting CMA [3-5]. Actually there are several published protocols with varied schemes of build-up and induction phases [6-8]. Additionally, different approaches (oral, sublingual, sublingual-oral) and durations until maintenance phase have been used [9, 10]. No protocol has already been gathered consensus. Overall, CM-OIT protocols include 2 distinct stages: an induction phase (progressive increases of threshold doses up to target dose) and a maintenance phase. The main goal is to achieve a long-lasting protective effect, which is assumed to be a state of clinical tolerance. Once achieved the maintenance phase patients are advised to keep regular intake of CM to ensure a tolerant status. Some patients can achieve sustained unresponsiveness, despite periods of withdrawal, while others need to fulfill regular consumption to maintain it. There is insufficient evidence whether clinical tolerance is transient or persistent [11]. Therefore, we cannot predict the risk of irregular CM ingestion. Some studies have reported loss of tolerance when ingestion is discontinued even on short periods [12, 13]. However, the minimal frequency of exposure to keep protection has not yet been established. For instance, a twice weekly ingestion scheme showed to be as effective as daily maintenance [14]. Clinical experience concerning long-term follow-up of these patients is limited.

In this survey, we aimed to assess the real life long-term efficacy and safety of the maintenance phase of a CM-OIT protocol that has been developed in our allergy center.

We envisioned evaluating the adherence to the protocol, defined as daily consumption of CM and free diet, along with the occurrence of allergic reactions. Furthermore, we sought to examine the course of maintenance phase in high-risk patients, namely with asthma and previous history of anaphylaxis.

MATERIALS AND METHODS

Study design

We have performed a prospective and noncontrolled study of all children and adolescents, who underwent CM-OIT and were subsequently followed at our department on maintenance dose (200-mL CM daily), for at least 36 months after reaching the maintenance phase (from 2009 until 2016).

Patients

Children aged 2–18 years, with persistent and moderate to severe IgE mediated CMA diagnosis were considered eligible for CM-OIT. CMA diagnosis was based on clinical history and an IgE-mediated underneath mechanism was confirmed by the presence of positive skin prick tests (wheal greater than or equal to 3 mm for CM and CM proteins) and/or

positive CM-specific IgE (greater than or equal to 0.35 kU/L). Serum-specific IgE antibodies (ImmunoCAP, Thermo Fisher Scientific, Waltham, MA, USA) to CM and milk proteins were performed before and after protocol. Before starting CM-OIT all children had a positive oral food challenge after an avoidance period or had a reproducible history on the previous year of an allergic reaction after exposure to small amounts of CM during the avoidance period.

Coexisting respiratory and cutaneous allergic diseases (allergic rhinitis, asthma, and atopic dermatitis) were properly controlled before the beginning of the CM-OIT protocol and were kept under surveillance during the maintenance period.

A trained medical team who designed the protocol and is experienced in this methodology has performed the overall clinical and social evaluation of all patients. Additionally, a systematic laboratory plan for follow-up was settled up during maintenance phase of CM-OIT protocol.

CM-OIT protocol

The induction phase comprises the introduction of increasing amounts of nondiluted, unprocessed, and unheated CM, beginning with sublingual doses and gradual increases of the threshold dose at predetermined intervals, within 14 to 28 days. The up dosing was always performed at our center. In each hospital session a fixed dose for daily ingestion to be taken at home has been established. The protocol (Table 1) includes usually 5 sessions (adjustable in case of a slower up-dosing regimen) [15]. To all patients an emergency written plan has been given, including oral, inhaled and injectable medication (adrenaline) to be used according to the severity of the allergic reaction. A mobile phone number of the medical team has been available for all patients, 24 hours a day.

The protocol has successfully if the target dose of 200 mL daily intake has been reached (corresponding to 6.6–7 g of CM proteins).

All legal guardians were fully informed about the procedures (risks and possible adverse reactions) and all of them signed an informed consent according to the Helsinki Declaration.

Statistical analysis

Data was analyzed using IBM SPSS Statistics ver. 23 (IBM Corp., Armonk, NY, USA). Quantitative variables with normal distribution were presented as mean ± standard deviation. Variables without normal distribution were presented as median and range. Categorical variables were described as absolute and relative frequencies and were expressed

Table 1. Dosing schedule of the induction phase of our cow's milk oral immunotherapy protocol

Variable	Session				
	1 (day 1)	2 (days 14 to 28)	3 (days 28 to 56)	4 (days 42 to 70)	5 (days 56 to 84)
Dose	1 Drop*	1 mL	10 mL	50 mL	100 mL
	2 Drops*	5 mL	10 mL	50 mL	100 mL
	3 Drops*	5 mL	20 mL	(interval 2 hr)	(interval 2 hr)
	4 Drops*	10 mL	20 mL	100 mL	200 mL
	0.1 mL	10 mL	50 mL		
	0.2 mL				
	0.5 mL				
	0.5 mL				
	1 mL				
Interval between doses	20–30 min (*sublingual)			20–120 min	
Maintenance dose (home)	0.5 to 1 mL twice daily	5 to 10 mL twice daily	20 to 50 mL twice daily	100 mL twice daily	200 mL daily (... progressive free diet)

as percentages. A *p* value less than 0.05 was considered as significant. Odds ratios were calculated with confidence interval of 95%. Associations between variables were determined using Pearson chi-square test, Mann-Whitney test and Fisher test.

RESULTS

Patients

We enrolled 42 patients (Table 2) who successfully undertaken CM-OIT protocol, defined as clinical tolerance of target dose (200 mL daily), with a follow-up time after reaching the maintenance phase for more than 36 months.

Almost all patients had other allergic diseases (95%) with significant prevalence of asthma (57%). Only 2 patients did not suffer from other allergic comorbidities (Table 2).

Before starting CM-OIT all children had proven CMA. Most of them have a reproducible history during last year of an allergic reaction after exposure to small amounts of CM during the avoidance period. We point out that 15 children (36%) had history of CM anaphylaxis. In those cases where there was not any history of accidental contact on the previous year an oral challenge test before starting CM-OIT was performed, which has been positive in all cases, from only after sublingual contact to less than 5 mL of CM.

Clinical outcomes

The median time of follow-up during the maintenance phase was 5.75 years (range, 3.25–8.75 years). Regarding the adherence to protocol during this period, 92% maintained diet without restrictions including daily ingestion of 200 mL of CM (36 of 39 adherent patients). Overall, 93% were adherent patients (39 of 42), since they keep daily ingestion of 200-mL CM.

Six patients (14%) had transient interruptions of daily ingestion; 2 due to allergic reactions, 3 due to poor adherence to the protocol, and 1 by personal choice.

Three patients completely suspended CM intake (noncompliance). After ingestion withdrawal, 2 of them developed allergic reactions, with progressively smaller amounts of CM. One patient kept butter intake, but has reported mucocutaneous symptoms up

Table 2. Clinical characteristics of the study group (n = 42)

Characteristic	Value
Male sex	60%
Age of CMA diagnosis (mo), median (range)	4 (0.5–12)
History of anaphylaxis	36%
Atopic dermatitis	33%
Asthma	57%
Allergic rhinitis	79%
Multiple food allergy (egg, tree nuts, peanut, fish, and shellfish)	24%
CM-OIT (induction phase)	
Age at CM-OIT initiation (yr), median (range)	6 (2–18)
Period until target dose (mo), mean ± SD	5.1 ± 2.7
CM-OIT (maintenance phase)	
Age at beginning (yr), median (range)	6.9 (2–18)
Age at last clinical evaluation (yr), median (range)	13 (6.7–23.8)
Time of follow-up (mo), median (range)	67 (37–103)

CMA, cow's milk allergy; CM-OIT, cow's milk-oral immunotherapy; SD, standard deviation.

Table 3. Safety of oral immunotherapy during maintenance

Allergic reaction & symptom	No.
Allergic reactions	
<3 episodes	16
≥3 episodes	3
No allergic reactions	23
Symptoms	
Mucocutaneous	17
Respiratory	7
Gastrointestinal	3
Systemic reactions (at least 2 systems involved)	2
Headaches	1

on exposure to processed foods containing higher amounts of CM. Another child had mucocutaneous and gastrointestinal symptoms after CM intake, for which he returned to soymilk. Nevertheless, he still tolerates processed foods with small amounts of baked CM.

During maintenance phase 45% (n = 19) developed allergic adverse reactions. Despite daily intake (n = 37), 41% (n = 15) had symptoms at some moment during the follow-up period. Overall, adherent patients with reactions during maintenance phase had a total of 33 symptomatic days in 67.5 months of mean follow-up time (Table 3).

CM-OIT elicited adverse reactions in 19 children who successfully completed the treatment. Cutaneous, oropharyngeal, and gastrointestinal symptoms were the most predominantly reported, mainly mild reactions. Six patients had moderate to severe systemic reactions. The occurrence of symptoms, even severe reactions, did not lead to discontinuation of CM-OIT. Further characterization of adverse reactions is detailed in Fig. 1.

Most of these reactions (n = 13) occurred in the presence of cofactors, such as exercise (n = 8) or during infections (acute rhinosinusitis-2, gastroenteritis-2, or respiratory infections-4). Few patients (7%) developed more than 3 adverse reaction episodes. All episodes had complete resolution at home after proper treatment. There were 2 cases of CM dependent exercise-induced anaphylaxis. Despite the occurrence of these 2 systemic reactions, adrenaline was not

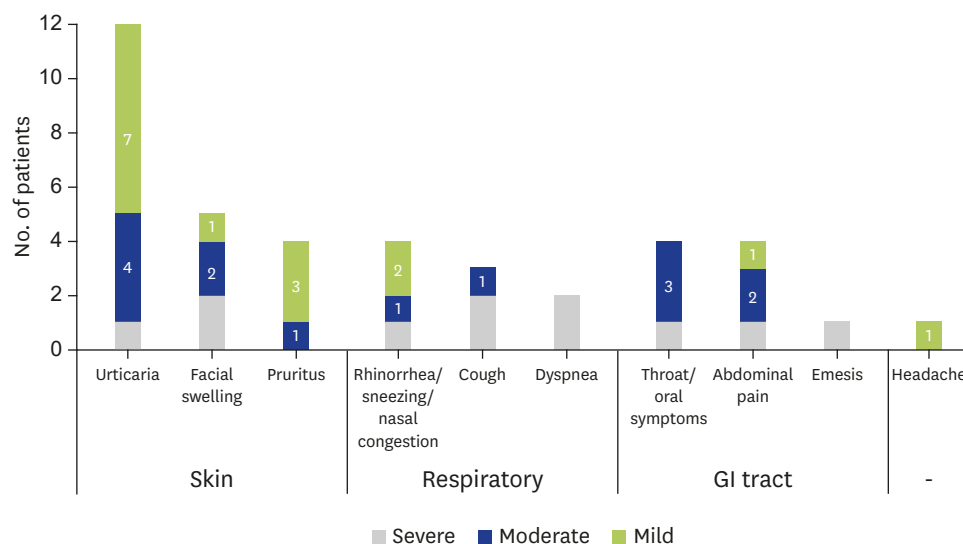


Fig. 1. Characterization of the adverse reactions: symptom grading. GI, gastrointestinal.

Table 4. Specific IgE levels and allergic adverse reactions before and after treatment.

Patient	Mean cow's milk IgE levels (kU/L)	
	Before OIT	After OIT
All patients (n = 42)	13.39	4.18
Patients with adverse reactions after OIT (n = 19)	13.34	4.99
Patients without adverse reactions after OIT (n = 23)	13.43	3.47

OIT, oral immunotherapy.

required to treat any of these patients. Those patients were successfully treated by caregivers using oral and inhaled medications.

Clinical risk factors

Since 45% of patients had reactions during maintenance, we have analyzed which clinical factors could be associated to the occurrence of allergic reactions. We have included a significant number of high-risk patients: 57% of asthmatics, 36% with previous history of anaphylaxis, and 21% with both risk factors (9 patients had asthma and history of anaphylaxis).

Among patients with history of anaphylaxis (15 patients), we found that 12 of them had reactions during the maintenance phase. A positive correlation, and highly statistically relevant, has been found between the occurrence of allergic reactions during maintenance phase and history of anaphylaxis to CM (80% of reactions in the group with anaphylaxis comparing to 26% without anaphylaxis history; odds ratio [OR], 11.382; $p < 0.001$).

Asthma history was slightly more frequent in children with reactions during the maintenance phase (58%) although not statistically significant ($p = 0.929$). In children with history of anaphylaxis, the coexistence of asthma was a risk factor, considered statistically significant, for the occurrence of reactions during the maintenance phase (37%; OR, 4.896; $p = 0.027$).

Comparing patients with and without adverse reactions (19 and 23 of them, respectively), we found no statistically significant difference in the occurrence of allergic reactions during the maintenance based on IgE levels to CM prior to OIT ($p = 0.4950$, Mann-Whitney). Even if we define a cut off value of 3 and 3.5 kU/L of CM-IgE levels we found the same result (no statistically significant; $p = 0.3532$ and 1.0000 , Fisher test). According to our results, specific IgE levels cannot be considered as risk factor of poor outcome (Table 4).

DISCUSSION

This real-life study sustains long-term efficacy and safety of CM-OIT. Our results have shown a great adherence to the protocol during the maintenance phase (93% of adherent patients). Daily intake of 200 mL ensures desensitization and enables diet without restrictions. However, among patients who fulfilled regular ingestion, 45% occasionally developed mild to moderate allergic reactions and 2 severe systemic reactions, which were exercise-induced. Symptoms were always successfully managed at home, according to an emergency written plan, under instructions provided by the medical team and by phone. The treatment of allergic reactions during the maintenance phase was the same as during the induction phase.

Regarding the safety, OIT studies for CM allergy has been associated an increased risk of systemic adverse reactions, as well as local (minor cutaneous/oropharyngeal/gastrointestinal) adverse reactions [16].

The safety of CM-OIT and the persistence of clinical tolerance to CM during maintenance phase depend on several factors: constant CM formulation; frequency of ingestion; control of cofactors and the risk factors; recognition and proper treatment of allergic reactions.

The CM formulation used during maintenance phase is the same UHT (ultrahigh temperature) pasteurized CM, with or without lactose, used during the induction phase. Other dairy products can be used, taking into consideration their protein CM content, to ensure that the daily dose of protein is progressively increased. We are strongly against the consumption of dairy product made from goat's or sheep's milk, for which we have given this recommendation to all patients. In our center, we have already had 2 anaphylactic reactions requiring intramuscular adrenaline during the maintenance phase (one on the first year and another on the fifth year of follow-up) due to the consumption of goat's cheese and goat's milk in 2 patients that otherwise were tolerant to CM daily ingestion and free diet. This event was also supported by a Spanish case report [17]. Daily CM dose should also be administered at room temperature, without heating, due to the possibility of reaction to the thermolabile conformational changes in CM proteins. Again, we have already noticed symptoms after consumption of heated CM, during maintenance phase.

Reactions during the maintenance phase might be related to poor adherence to the daily ingestion [18, 19] or related to the effect of cofactors [20].

Patients who discontinued CM ingestion, lost clinical tolerance even to small amounts of CM. Overall, in our study, four patients discontinued CM daily ingestion (noncompliance): one patient still tolerates butter, but reacts to other dairy products containing higher amounts of CM protein; another kept consumption of 1 yogurt weekly or less and the remaining two patients developed allergic reactions with small amounts of CM, tolerating low content of baked milk in processed food. It is likely that the efficacy of CM-OIT only lasts if CM is regularly consumed. While some patients might achieve CM sustained unresponsiveness, others maintain tolerance only if they keep regular ingestion, though not daily [18, 19].

Almost all protocols of CM-OIT recommend administering daily doses during the maintenance phase. There is no strong evidence to recommend less frequent dosing, since a lesser frequency of exposure may result in losing the desensitization effect [21]. No differences in the occurrence of allergic reactions were observed comparing protocol schedules involving daily doses to twice weekly doses [14]. However, a decrease in dosing frequency or lack of adherence to daily ingestion might result in higher rate of allergic reactions [18].

Concerning long-term efficacy, the first long-term follow-up study (4 years and 8 months) performed by Meglio et al. [19] confirmed a persistent effect of oral desensitization protocol, with 13 out of 14 children showing total tolerance. Lately, other authors reported less satisfactory outcomes. A Spanish prospective study [8] that evaluated the efficacy of a rush desensitization protocol in 18 CMA children found out that after 2 years 72% of them achieved full tolerance, keeping a diet without restrictions. Looking at the persistence of allergic reactions, a recent multicentric Italian study [22] showed that some children (36.4%) who consumed the offending food occasionally, developed reactions similarly to those which reacted before the intervention. According to Keet et al. [23], children followed-up after 3.2 to 4.5 years after CM-OIT, lost desensitization over time, and only 31% of them tolerated CM with minimal or without symptoms. Elizur et al. [24] reported that among 92.3% (180 of

195) who were consuming milk protein regularly, half of them experienced adverse reactions, mostly mild. Nevertheless 6.7% required injectable adrenaline.

The presence of cofactors, such as exercise following CM ingestion, fasting, infection, or uncontrolled asthma are risk factors for the occurrence of allergic reactions even in the maintenance phase [18, 20, 25-27]. Considering the influence of these cofactors not only during induction phase but also during maintenance, patients are strictly advised to avoid vigorous physical activity within 2 hours after CM ingestion. In our experience, some patients have reported mild to moderate symptoms when they had gastrointestinal or respiratory infections. These infections explain a decrease in the clinical threshold of CM, and therefore it is advised to have a transient reduction of the daily dose of CM until complete recovery. When the dosing frequency was reduced or temporarily suspended for a few days, the administration of the following dose has been made at hospital, under medical supervision, due to a higher risk of an allergic reaction. This up-dosing session at hospital setting was well-tolerated in all cases. Considering the remaining cofactors, exercise has assumed to be the major trigger to elicit allergic reactions in our patients (62%). We have observed that some patients developed reactions when CM ingestion was immediately followed by exercise (i.e., running, football, volleyball). This association between exercise and breakdown in clinical tolerance to CM has been described [26, 28]. The underlying pathophysiological mechanisms have not yet been clearly understood. One of the hypotheses proposed is related to the induction of a higher gastrointestinal permeability during exercise [29]. Although there is no evidence of the effect of fasting upon the safety of CM-OIT, most studies recommend avoiding administration of the dose during fasting, since there is a faster allergen absorption and an increased risk of allergic reactions [21].

We have identified the history of previous anaphylaxis as a risk factor for the occurrence of mild to moderate reactions during the maintenance period. Comparing to other studies, which included children with severe CMA (anaphylaxis), the incidence of adverse reactions during the rush phase and at home was higher than was observed in other studies where high-risk patients were excluded [6, 7]. A higher reaction rate after immunotherapy was associated with the occurrence of more anaphylactic episodes before treatment and at a lower starting dose [24].

In our asthmatic patients, the occurrence of allergic reactions during maintenance was slightly higher, but this observation was not statistically significant. Several studies reported that coexisting asthma is a risk factor for the persistence of IgE-mediated CMA [2, 30]. In this context, Vázquez-Ortiz et al. [20] demonstrated that patients with asthma have more frequent and persistent reactions during CM-OIT. A controlled study including patients with and without asthma showed that asthmatic patients are at risk of severe reactions during CM-OIT and are less likely to reach complete desensitization. Nonetheless in most cases, the reactions subside with time [31]. In patients with previous anaphylaxis, the coexistence of asthma is a risk factor associated to fatal anaphylactic reactions and asthma is the most important risk factor in CM-OIT, causing more severe and persistent allergic reactions during treatment [21].

In our patients, CM-IgE antibodies were not helpful as predictive risk factors of adverse reactions. The majority of the studies reported that OIT did not reduce allergen-specific IgE levels [16]. However, some authors found that children who discontinued OIT, due to adverse effects, had IgE antibodies that bound to CM peptides with greater intensity, broader

diversity, and greater affinity than in children who successfully completed OIT [32] as well as high IgE levels to milk proteins are associated with lower maintenance dose reached [33].

Few studies have assessed if OIT can lead to permanent tolerance or if they have only a transient desensitization effect. There are few studies on the long-term evolution and no evidence has yet been obtained regarding the minimum length of the maintenance phase. Some patients might lose tolerance after short periods of CM ingestion interruption [11]. Overall, in the literature the length of follow-up ranges from 3 up to 6 years and the desensitization is maintained from 31% to 100% of the patients [21]. According to Meglio et al. [19], one child interrupted the consumption of milk for 1 month and went back to lower dose (100 mL) straightforward. A Spanish prospective study has reported a worst outcome [8], in which 1 patient discontinued CM at home, after having reached maintenance, and developed an anaphylactic reaction at the hospital during an attempt to reintroduce it again. In a controlled study, 40% of children (6 of 15) who overcome a CM challenge after a 60-week maintenance period, regained reactivity after CM avoidance, 2 of them only 1 week afterwards and the others after 6 weeks [13]. Sato et al. [11] demonstrated that after a 2-year follow-up (daily intake of 200 mL) followed by an interruption for 2 weeks of CM ingestion, 75% of patients had been successfully desensitized, proven by an open oral challenge. The long-term outcomes of CM-OIT are heterogeneous: some patients lose their desensitization status over a long period of time; others maintain the tolerant status and can continue to consume a full serving or lower doses, without symptoms [18, 23].

Considering our results, along with published data, the benefits of a CM-OIT protocol seems to be quickly lost after its suspension. It is likely that the protective effect of an OIT can eventually require long-term ongoing exposure to develop tolerance. We can speculate that daily ingestion keeps a stable threshold to avoid allergic reactions. If daily intake is interrupted, the protective threshold decreases and might be progressively lost over time.

This real-life study showed that long-term course and outcomes are unpredictable and heterogeneous, depending on clinical risk factors (previous anaphylaxis and asthma), protocol adherence and cofactors. A significant number of patients still react occasionally during maintenance phase. Daily CM ingestion of the full serving amount (200 mL) enables a diet without restrictions, ensuring the maintenance of CM desensitization and decreasing the chance of a severe reaction upon accidental exposure. This intervention had a positive impact on both children and their families. We consider crucial to keep these patients under regular follow-up, emphasizing the importance of daily intake to guarantee the safety of the treatment, as well as the awareness of active disease and the possibility of the occurrence of allergic reactions.

REFERENCES

1. Nwaru BI, Hickstein L, Panesar SS, Roberts G, Muraro A, Sheikh AEAACI Food Allergy and Anaphylaxis Guidelines Group. Prevalence of common food allergies in Europe: a systematic review and meta-analysis. *Allergy* 2014;69:992-1007.
[PUBMED](#) | [CROSSREF](#)
2. Skripak JM, Matsui EC, Mudd K, Wood RA. The natural history of IgE-mediated cow's milk allergy. *J Allergy Clin Immunol* 2007;120:1172-7.
[PUBMED](#) | [CROSSREF](#)

3. Yeung JP, Kloda LA, McDevitt J, Ben-Shoshan M, Alizadehfar R. Oral immunotherapy for milk allergy. *Cochrane Database Syst Rev* 2012;11:CD009542.
[PUBMED](#)
4. Martorell Calatayud C, Muriel García A, Martorell Aragonés A, De La Hoz Caballer B. Safety and efficacy profile and immunological changes associated with oral immunotherapy for IgE-mediated cow's milk allergy in children: systematic review and meta-analysis. *J Investig Allergol Clin Immunol* 2014;24:298-307.
[PUBMED](#)
5. Santos N, Gaspar A, Borrego LM, Morais-Almeida M. Successful oral tolerance induction to cow's milk in a child with allergy to extensively hydrolysed formula. *Allergol Immunopathol (Madr)* 2015;43:216-8.
[PUBMED](#) | [CROSSREF](#)
6. Longo G, Barbi E, Berti I, Meneghetti R, Pittalis A, Ronfani L, Ventura A. Specific oral tolerance induction in children with very severe cow's milk-induced reactions. *J Allergy Clin Immunol* 2008;121:343-7.
[PUBMED](#) | [CROSSREF](#)
7. Pajno GB, Caminiti L, Ruggeri P, De Luca R, Vita D, La Rosa M, Passalacqua G. Oral immunotherapy for cow's milk allergy with a weekly up-dosing regimen: a randomized single-blind controlled study. *Ann Allergy Asthma Immunol* 2010;105:376-81.
[PUBMED](#) | [CROSSREF](#)
8. González Jiménez D, Larrea Tamayo E, Díaz Martin JJ, Molinos Norniella C, Pérez Solis D, Menéndez Arias C, Jiménez Treviño S, Bousoño García C. Oral rush desensitization for cow milk allergy: clinical and immunological follow-up. *An Pediatr (Barc)* 2013;79:346-51.
[PUBMED](#)
9. Pajno GB, Cox L, Caminiti L, Ramistella V, Crisafulli G. Oral Immunotherapy for treatment of immunoglobulin E-mediated food allergy: the transition to clinical practice. *Pediatr Allergy Immunol Pulmonol* 2014;27:42-50.
[PUBMED](#) | [CROSSREF](#)
10. Passalacqua G, Landi M, Pajno GB. Oral immunotherapy for cow's milk allergy. *Curr Opin Allergy Clin Immunol* 2012;12:271-7.
[PUBMED](#) | [CROSSREF](#)
11. Sato S, Yanagida N, Ogura K, Asami T, Okada Y, Koike Y, Iikura K, Syukuya A, Ebisawa M. Immunotherapy in food allergy: towards new strategies. *Asian Pac J Allergy Immunol* 2014;32:195-202.
[PUBMED](#)
12. Rolinck-Werninghaus C, Staden U, Mehl A, Hamelmann E, Beyer K, Niggemann B. Specific oral tolerance induction with food in children: transient or persistent effect on food allergy? *Allergy* 2005;60:1320-2.
[PUBMED](#) | [CROSSREF](#)
13. Keet CA, Frischmeyer-Guerrero PA, Thyagarajan A, Schroeder JT, Hamilton RG, Boden S, Steele P, Driggers S, Burks AW, Wood RA. The safety and efficacy of sublingual and oral immunotherapy for milk allergy. *J Allergy Clin Immunol* 2012;129:448-55, 455.e1-5.
[PUBMED](#) | [CROSSREF](#)
14. Pajno GB, Caminiti L, Salzano G, Crisafulli G, Aversa T, Messina MF, Wasniewska M, Passalacqua G. Comparison between two maintenance feeding regimens after successful cow's milk oral desensitization. *Pediatr Allergy Immunol* 2013;24:376-81.
[PUBMED](#) | [CROSSREF](#)
15. Morais-Almeida M, Piedade S, Couto M, Sampaio G, Santa-Marta C, Gaspar A. Innovation in specific oral tolerance induction in children with anaphylaxis to cow's milk proteins. *Rev Port Imunoalergologia* 2011;19:161-9.
16. Nurmatov U, Devereux G, Worth A, Healy L, Sheikh A. Effectiveness and safety of orally administered immunotherapy for food allergies: a systematic review and meta-analysis. *Br J Nutr* 2014;111:12-22.
[PUBMED](#) | [CROSSREF](#)
17. Alonso-Lebrero E, Fuentes V, Zapatero L, Pérez-Bustamante S, Pineda F, Martínez-Molero MI. Goat's milk allergies in children following specific oral tolerance induction to cow's milk. *Allergol Immunopathol (Madr)* 2008;36:180-1.
[PUBMED](#) | [CROSSREF](#)
18. Staden U, Rolinck-Werninghaus C, Brewe F, Wahn U, Niggemann B, Beyer K. Specific oral tolerance induction in food allergy in children: efficacy and clinical patterns of reaction. *Allergy* 2007;62:1261-9.
[PUBMED](#) | [CROSSREF](#)
19. Meglio P, Giampietro PG, Gianni S, Galli E. Oral desensitization in children with immunoglobulin E-mediated cow's milk allergy--follow-up at 4 yr and 8 months. *Pediatr Allergy Immunol* 2008;19:412-9.
[PUBMED](#) | [CROSSREF](#)

20. Vázquez-Ortiz M, Alvaro-Lozano M, Alsina L, Garcia-Paba MB, Piquer-Gibert M, Giner-Muñoz MT, Lozano J, Domínguez-Sánchez O, Jiménez R, Dias M, Martín-Mateos MA, Plaza-Martín AM. Safety and predictors of adverse events during oral immunotherapy for milk allergy: severity of reaction at oral challenge, specific IgE and prick test. *Clin Exp Allergy* 2013;43:92-102.
[PUBMED](#) | [CROSSREF](#)
21. Martorell A, Alonso E, Echeverría L, Escudero C, Garcia-Rodríguez R, Blasco C, Bone J, Borja-Segade J, Bracamonte T, Claver A, Corzo JL, De la Hoz B, Del Olmo R, Dominguez O, Fuentes-Aparicio V, Guallar I, Larramona H, Martín-Muñoz F, Matheu V, Michavila A, Ojeda I, Ojeda P, Piquer M, Poza P, Reche M, Río PRD, Rodríguez M, Ruano F, Sánchez-García S, Terrados S, Valdesoiro L, Vazquez-Ortiz M. Expert panel selected from members of the Spanish Societies of Pediatric Allergology, Asthma and Clinical Immunology (SEICAP) and Allergology and Clinical Immunology (SEAC). Oral immunotherapy for food allergy: A Spanish guideline. Egg and milk immunotherapy Spanish guide (ITEMS GUIDE). Part 2: Maintenance phase of cow milk (CM) and egg oral immunotherapy (OIT), special treatment dosing schedules. Models of dosing schedules of OIT with CM and EGG. *Allergol Immunopathol (Madr)* 2017;45:508-18.
[PUBMED](#) | [CROSSREF](#)
22. Meglio P, Caminiti L, Pajno GB, Dello Iacono I, Tripodi S, Verga MC, Martelli ASIAIP Group. The oral food desensitization in the Italian allergy centres. *Eur Ann Allergy Clin Immunol* 2015;47:68-76.
[PUBMED](#)
23. Keet CA, Seopaul S, Knorr S, Narisety S, Skripak J, Wood RA. Long-term follow-up of oral immunotherapy for cow's milk allergy. *J Allergy Clin Immunol* 2013;132:737-739.e6.
[PUBMED](#) | [CROSSREF](#)
24. Elizur A, Appel MY, Goldberg MR, Yichie T, Levy MB, Nachshon L, Katz Y. Clinical and laboratory 2-year outcome of oral immunotherapy in patients with cow's milk allergy. *Allergy* 2016;71:275-8.
[PUBMED](#) | [CROSSREF](#)
25. Barbi E, Longo G, Berti I, Matarazzo L, Rubert L, Saccari A, Lenisa I, Ronfani L, Radillo O, Ventura A. Adverse effects during specific oral tolerance induction: in home phase. *Allergol Immunopathol (Madr)* 2012;40:41-50.
[PUBMED](#) | [CROSSREF](#)
26. Couto M, Gaspar A, Santa-Marta C, Morais-Almeida M. Cow's milk dependent exercise-induced urticaria after oral tolerance induction in an adolescent. *Allergol Immunopathol (Madr)* 2012;40:67-8.
[PUBMED](#) | [CROSSREF](#)
27. Piedade S, Morais-Almeida M. Indução de tolerância em alergia alimentar. In: Castro FF, Galvão CE, editors. *Imunoterapia*. São Paulo: Editora Manole Lda; 2011. p. 125-47.
28. Caminiti L, Passalacqua G, Vita D, Ruggeri P, Barberio G, Pajno GB. Food-exercise-induced anaphylaxis in a boy successfully desensitized to cow milk. *Allergy* 2007;62:335-6.
[PUBMED](#) | [CROSSREF](#)
29. Ansley L, Bonini M, Delgado L, Del Giacco S, Du Toit G, Khaitov M, Kurowski M, Hull JH, Moreira A, Robson-Ansley PJ. Pathophysiological mechanisms of exercise-induced anaphylaxis: an EAACI position statement. *Allergy* 2015;70:1212-21.
[PUBMED](#) | [CROSSREF](#)
30. Saarinen KM, Pelkonen AS, Mäkelä MJ, Savilahti E. Clinical course and prognosis of cow's milk allergy are dependent on milk-specific IgE status. *J Allergy Clin Immunol* 2005;116:869-75.
[PUBMED](#) | [CROSSREF](#)
31. Elizur A, Goldberg MR, Levy MB, Nachshon L, Katz Y. Oral immunotherapy in cow's milk allergic patients: course and long-term outcome according to asthma status. *Ann Allergy Asthma Immunol* 2015;114:240-4.e1.
[PUBMED](#) | [CROSSREF](#)
32. Savilahti EM, Kuitunen M, Valori M, Rantanen V, Bardina L, Gimenez G, Mäkelä MJ, Hautaniemi S, Savilahti E, Sampson HA. Use of IgE and IgG4 epitope binding to predict the outcome of oral immunotherapy in cow's milk allergy. *Pediatr Allergy Immunol* 2014;25:227-35.
[PUBMED](#) | [CROSSREF](#)
33. Kuitunen M, Englund H, Remes S, Movérare R, Pelkonen A, Borres MP, Mäkelä MJ. High IgE levels to α -lactalbumin, β -lactoglobulin and casein predict less successful cow's milk oral immunotherapy. *Allergy* 2015;70:955-62.
[PUBMED](#) | [CROSSREF](#)